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Operating Manual for the Bedford Institute of Oceanography Automated Dissolved Oxygen Titration System

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by

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92-05734/

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ABSTRACT

Jones, E.P., F. Zemlyak, and P. Stewart. 1992. Operating Manual for the Bedford Institute of Oceanography Automated Dissolved Oxygen Titration System. Can. Tech. Rep. of Hydrography and Ocean Sci. 138: iv + 51 p.

The Department of Fisheries and Oceans Physical and Chemical Sciences Branch at the Bedford Institute of Oceanography uses a computer-automated titration system developed in-house to assist in carrying out routine oxygen titrations by the Winkler method. The system consists of an IBM PC computer and commercially available expansion boards and modules, a Metrohm Multi-Dosimat 655 automatic titrator, a Brinkmann Model PC 800 Probe Colorimeter, and the 'OXYGEN' computer program. The computer controls the titrator, monitors transmittance of the sample as the measure of completeness of titration, determines endpoints, determines oxygen concentration and logs data. This publication describes how to use the system, outlines the oxygen determination method and gives working details of essential equipment.

RÉSUMÉ

Jones, E.P., F. Zemlyak, and P. Stewart. 1992. Operating Manual for the Bedford Institute of Oceanography Automated Dissolved Oxygen Titration System. Can. Tech. Rep. of Hydrography and Ocean Sci. 138: iv + 51 p.

La Direction des sciences physiques et chimiques du ministère des Pêches et des Océans a l'Institut océanographique de Bedford utilise un système informatisé de titrage conçu à l'Institut pour procéder aux opérations courantes de titrage de l'oxygène par la méthode Winkler. Ce système se compose d'un mini-ordinateur IBM, de cartes et de modules d'expansion en vente dans le commerce, d'un appareil de titrage automatique Metrohm Multi-Dosimat 655, d'un mini-ordinateur de colorimétrie de modèle Brinkman 800 et du logiciel "OXYGEN". L'ordinateur commande l'appareil de titrage, surveille la transmission de l'échantillon, indiquant l'aboutissement du processus de titrage, détermine les points extrêmes ainsi que la concentration d'oxygène et emmagasine des données. La présente publication explique comment utiliser le système et fournit des renseignements utiles sur son matériel essentiel.

1. INTRODUCTION

Oxygen is an essential element for life and one of the basic parameters used in oceanographic studies. The Department of Fisheries and Oceans, Physical and Chemical Sciences Branch at the Bedford Institute of Oceanography in Dartmouth, Nova Scotia, has routinely monitored oxygen in physical, chemical and biological studies of ocean waters using the modified Winkler titration and best available analytical techniques.

In the mid- to late 1970s electro-mechanical titrators came into use, simplifying repetitious tasks and improving precision in determinations as described in Levy *et al* (1977). The latest development, which is the subject of this manual, responds to the logical step to computer control and data logging, and instrument capabilities to communicate with computer systems. Computer control of the titration simplifies the oxygen analysis in a number of ways. It eliminates the need for an operator to determine the endpoint, and consequently dispenses with the use of starch indicator. Endpoint is determined solely by measurement of transmittance at 420 nanometers. The computer program also filters the data, carries out oxygen calculations instantly, and logs data. The present automated system using a personal computer was developed circa 1987 and has been used routinely in Department of Fisheries and Oceans oceanographic sampling activities.

This manual describes the automated titration system and operating procedures for the benefit of other institutions and internal users. Enough detail is presented to allow a user to assemble a complete system. The computer software for the oxygen titration system is available to all potential users.

2. DISSOLVED OXYGEN METHOD

2.1 Basics and Background

The automated procedure described in this publication is based on the oxygen determination method developed by Physical and Chemical Sciences Branch (PCS) of the Bedford Institute of Oceanography (BIO) (Levy *et al* 1977) and used routinely in scientific surveys carried out by BIO. The PCS procedure is a modified Winkler method based on Carritt and Carpenter (1966) and using a 'whole bottle' titration to eliminate errors due to pipetting in the widely-used Strickland and Parsons (1972) method. The present procedure eliminates the use of starch indicator, introduces the use of a wetting agent to reduce bubble formation on the optical probe used to measure transmittance of the sample, and employs dispensing equipment of higher precision and reliability. The method does not, however, accommodate changes in sample volume with temperature (e.g. Culberson, World Ocean Circulation Experiment (WOCE)), and significant errors can be introduced if the temperature of the sample at the time of collection is more than a few degrees different from the temperature at which the sample bottle was calibrated.

The following description of the chemical basis of the present modified Winkler titration is taken from Levy *et al* (1977). A precipitate of divalent manganous hydroxide is produced in a measured volume of seawater by adding a soluble manganous salt and a strong alkali. In this case a solution of sodium hydroxide and sodium iodide ('alkaline iodide'), and manganous chloride are used. The resulting white, manganous hydroxide precipitate scavenges oxygen from the sample and produces a trivalent brown hydroxide. Addition of concentrated sulfuric acid dissolves the hydroxide and releases the trivalent manganese ion, which reacts with iodide to form a brown iodine solution in which the amount of iodine is chemically equivalent to the amount of oxygen present. The iodine and excess iodide form a complex which can be titrated with a standardized thiosulfate solution (in this case sodium thiosulfate) to either a starch endpoint (iodine in the presence of starch gives an intense blue colour) or to the disappearance of the yellow-brown iodine colour, measured by transmittance using the automated method.

2.2 Reagents and Apparatus

Reagents

The following reagents are required for sampling:

Manganous chloride ($MnCl_2 \cdot 4H_20$)--600 g/L (3M)
Alkaline iodide-- NaOH, 320 g/L (8N), and NaI, 600 g/L (4M).

Analytical reagents are:

Sulfuric acid-- 280 mL/L of concentrated H_2SO_4 (10N)
Sodium thiosulfate-- 12.4 g/L (0.05N)
Potassium iodate (standard)-- 0.3567 g/L (0.01N)
Wetting Agent-- Technicon Agent A or equivalent.

The iodate standard is prepared by accurately weighing KIO_3 that has been dried to constant weight at 115° C and dissolved in exactly one litre of distilled water. Iodate standards can also be purchased commercially.

In the event of a failure of the automated system, the titration can be carried out manually but will require use of a starch indicator solution to determine endpoint. Starch indicator is usually prepared by making a starch slurry and adding it to boiling water to achieve a final concentration of about 1%.

Apparatus

Sample Bottles--The method uses 125 mL 'iodine bottles' with a custom ground glass stopper (Figure 1) as recommended in Levy *et al* (1977). Bottles and stoppers must have matching numbers and each set must have its volume determined before use.

Figure 1. 125 mL iodine bottle and stirring bar.

Reagent Dispenser--Use a standard laboratory dispenser capable of being used in an alkaline solution (Figure 2). Each dispenser should be attached to 15 cm of 1 mm I.D. polyethylene or teflon tubing to enable the reagents to be dispensed near the bottom of the sample flask. Wetting agent is added from an eye dropper or other convenient dispenser.

Acid Dispenser--The analysis requires an adjustable glass repipet of approximately 5 mL capacity with a 7.5 mm polyethylene or teflon dispensing tube (Figure 3).

Incidental Features--A short (2mm) piece of Tygon tubing is placed on each end of the 2.5 cm magnetic stirring bar to enable it to stay centred on the bottom of the oxygen sample bottle (Figure 1).

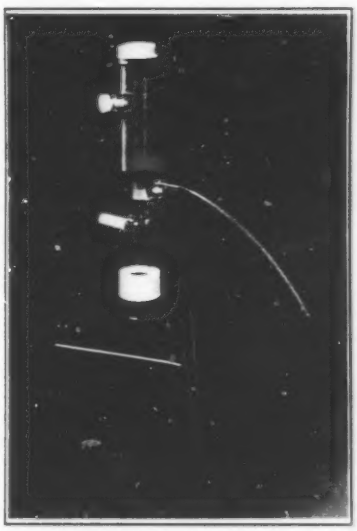

Figure 2. Alkaline iodide dispenser.

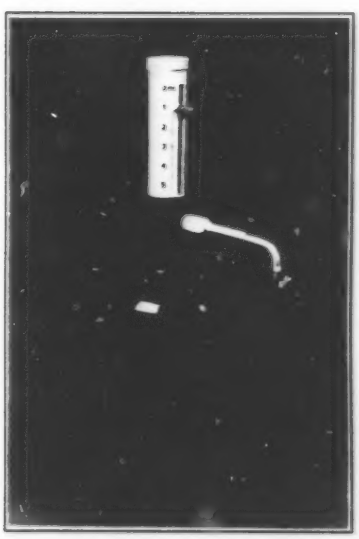

Figure 3. Acid dispenser.

2.3 Method

Sampling

The sampling technique described below ensures that the sample has minimal contact with the atmosphere. A 15-20 cm piece of latex or silicone rubber tubing is placed on the stopcock of the Niskin water sampler. Water flowing from the sampler is used to rinse out the oxygen flask first by being directed into the inverted flask and allowed to flow from top to bottom in a circular fashion. Then, while pinching the tube to restrict flow, the flask is righted and filled and flow is controlled to eliminate bubble formation. An overflow of two to three flask volumes helps ensure an uncontaminated sample, and provides an opportunity to rinse the stopper. Immediately, 1 mL each of the alkaline iodide and manganous chloride reagents are injected into the sample, one after the other (the order is not important). The dispensing tubes should be immersed about 1 cm into the sample before injecting the reagents. After the chemicals have been added, the stopper is replaced without trapping bubbles, and the flask is shaken to mix the reagents. Shaking should be repeated once the precipitate has settled to the lower third of the flask, and the precipitate should be allowed to settle again before analysis. Samples should be analyzed as soon as possible after sampling.

Sample Titration

The stopper is withdrawn from the sample flask and 1.5 mL of sulphuric acid is injected just above the level of the precipitate, using the polyethylene or teflon dispensing tube. The stopper is replaced and the sample shaken to dissolve the precipitate. Then the stopper is removed, residual sample is washed into the flask using distilled water (Note: More acid may be required if the precipitate does not completely disappear. Dissolved oxygen in the rinse water will not affect the oxygen measurement.), and one drop of wetting agent and the magnetic stirring bar are added. Magnetic stirring bars must be rinsed with distilled water between titrations. Titration begins immediately as the iodine in solution is volatile and delay can result in lowered oxygen measurements.

The automated titration is described in Section 4. If the titration must be completed manually, titrate to a pale yellow colour, add 1 mL of the starch indicator solution and titrate until the solution loses its blue colour. Test samples for checking performance of the automated system may be prepared by using the above procedure with distilled water, or seawater samples.

Standards and Blanks

Standards--The standardization is similar to that presented in Levy *et al* (1977) except that 1.5 mL of sulfuric acid reagent instead of 1.0 mL, and a drop of wetting agent is added. Ten (10.00) mL of 0.01 N potassium iodate are pipetted into a clean sample flask which is then almost filled with distilled water. The sulfuric acid and 1 mL of alkaline iodide solution are added and the solution is titrated in triplicate with the stock sodium thiosulfate solution.

Blanks--One millilitre of 0.01 N potassium iodate is pipetted into a clean sample flask which is then nearly filled with distilled water. Sulfuric acid reagent (1.5 mL) and 1 mL of alkaline iodide reagent are added and the sample mixed thoroughly. Then 1 mL of manganous chloride reagent and a drop of wetting agent are added. After this sample has been titrated, a second 1 mL of 0.01N potassium iodate is added and the titration is repeated. The difference between first and second titrations is the reagent blank and may be positive or negative. The blank titration is done in triplicate. It is not necessary to manually record titrant volumes during blank titrations.

2.4 Oxygen Calculation

The oxygen concentration from Levy *et al* (1977) is:

$$DO\ (mL/L) = \frac{(R_{sam} - R_{blk}) \times V_{IO3} \times N_{IO3} \times 5{,}598\ mL\ O_2/equivalent}{(R_{std} - R_{blk})\ (V_{bot} - V_{reg})} - DO_{reg}$$

where
- R_{sam} = volume of titrant for sample
- R_{blk} = volume of titrant for blank
- R_{std} = volume of titrant for standard
- V_{IO3} = volume of KIO3 standard (mL) = 10 mL
- V_{bot} = volume of sample bottle (mL)
- V_{reg} = volume of sample displaced by the manganous and alkaline iodide reagents (mL)
- N_{IO3} = normality of KIO3 standard (eq/L) = 0.01
- DO_{reg} = oxygen added with reagents = 0.018

For values of reagents used here, the equation reduces to:

$$DO\ (mL/L) = \frac{(R_{sam} - R_{blk}) \times 559.8}{(R_{std} - R_{blk})\ (V_{bot} - 2)} - 0.018$$

Culbertson (in prep.) suggests that the value for DO_{reg} should be 0.0017 following Murray *et al* (1968). This value is not used in the procedure at BIO but may be incorporated in future.

3. FIRST TIME SET-UP

3.1 Overview

The automated oxygen titration system consists of an IBM PC computer linked to a Brinkmann PC800 colorimeter and a Metrohm 655 Multi-Dosimat Automatic Titrator (Figure 4). The computer operates the titrator; monitors the progression of the titration by transmittance measurements from a colorimeter probe in the sample, monitors volume of titrant dispensed, detects and computes endpoints, and manages a database containing results and data used in determining oxygen values.

The instructions in this section enable a user to assemble the components of the system. Detailed instructions on building and modifying off-the-shelf components for use in the system are presented in Section 5.

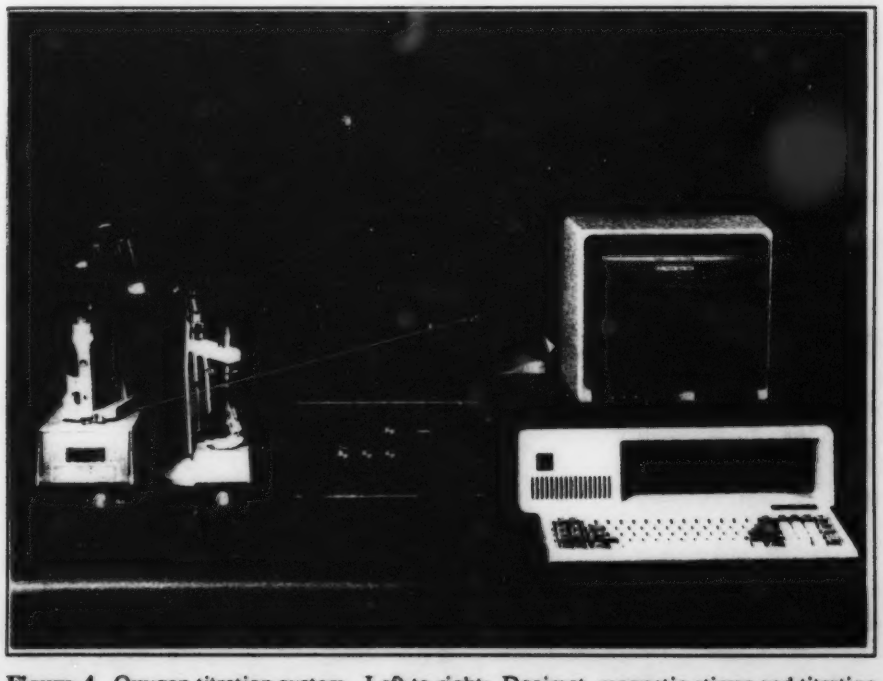

Figure 4. Oxygen titration system. Left to right: Dosimat, magnetic stirrer and titration flask, colorimeter, and computer.

3.2 Setting Up the Computer and Printer

The IBM PC compatible computer used in the system has two floppy disk drives (A: and B:) and an expansion board to handle input and output from the instrumentation. Minor program modifications can be made to allow other configurations. Assembly of the expansion board is described in Section 5.

Use normal precautions for an electrical device to avoid shocks and jarring in the set-up of the personal computer. Connect keyboard, monitor and printer using appropriate cables. The printer cable is the only one you will have to connect to the back of the computer. Four strap-type cables come out of the back of the computer and originate directly from the expansion board inside. One connects to the colorimeter, two to the titrator, and a fourth is an RS-232C connector. These will be connected later. Make sure all connections are tight and verify that the printer is turned on and that the connection is OK. (Note: Under certain circumstances if there is a poor printer connection or if the printer is not connected or is not on line, some of the features in the automatic titration program will not work).

Turn on the computer, monitor, and printer to ensure that they are working properly. Since the computer does not have an internal disk or 'hard' drive, the diskette provided with the oxygen titration system must be inserted in the A: drive and the drive door must be closed for the system to start properly. The IBM PC computer will normally sit briefly after it is turned on and before it begins running the start-up or 'boot' program. The system will ask for the date and time. Verify that the system displays the prompt A:\>, indicating that it is functioning properly.

3.3 Setting up the Colorimeter

Attach the fibre optic probe to the colorimeter. The probe is stainless steel and has a rectangular opening or sampling region near the end, on the bottom of which is a small mirror. The light guide or cable connecting the probe to the instrument has two leads which plug into the face of the instrument--one for the source beam and one for the return. Each socket has a guide pin which permits only the correct lead to be inserted. Fix the plugs in place by tightening the thumb screws. Occasionally the screws may become loose and affect the fibre optic connection, resulting in bad readings, so it is wise to check them from time to time. The return has an on-line optical filter mounted where the line enters the colorimeter. Check to ensure that the filter is in place and that it is the correct wavelength (420 nanometers). Never turn the instrument on without a filter in place.

Insert the probe into the holder mounted on the articulated arm stand (Figure 5). The probe occupies the larger of two holes in a teflon block which position it next to the tube through which the titrant is dispensed. Clean the optical surfaces periodically with a cotton swab or 'Q-tip'.

Connect the appropriate cable from the computer to the rear of the colorimeter. Only one of the cables leaving the computer will match this plug which is a Centronics-type connector. Plug in the colorimeter to a 110 V AC, grounded electrical outlet, and allow the instrument to warm up for at least five minutes--longer if it is much below room temperature.

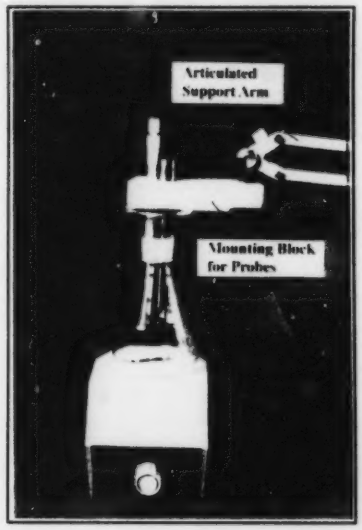

Figure 5. Fibre optic probe and dispensing tip mounted on articulated arm.

Check lamp voltage by turning the 'Function' knob to 'Volt'. Lamp voltage must be 7.5 to 8 Volts for proper functioning of the instrument. (Note: problems with the system have occasionally been traced to inadequate lamp voltage). Unplug one of the leads from the light guide and adjust the transmittance to zero if necessary using the slotted knob on the rear of the instrument.

Replace the lead from the probe, and immerse the probe tip in distilled water. Adjust the transmittance to about 100%. Note that under normal operations it is only necessary to check transmittance occasionally. Since the oxygen determination uses differences in transmittance and not absolute values, variations during a survey or analysis session will not affect oxygen concentrations determined by the system. Transmittance occasionally will read >100%.

3.4 Setting up the Titrator

Plug the Metrohm 655 Multi-Dosimat (Dosimat) minus the 'snap-in burette unit' platform into a standard 110 V AC grounded electrical outlet and turn it on using the button at the back of the instrument. The Dosimat doesn't need a warm-up. Don't hook up the cables to the computer yet as this disables internal functions of the system which are necessary for setup. On start-up, the instrument display will read '88.88'.

The 'snap-in burette unit' slides into position on the Dosimat. The burette is held in alignment by a positioning collar and two thumb screws. The shaft of the burette plunger

extends beneath the 'snap-in burette unit' where it links by means of a metal pin to a shaft on the Dosimat. You must align it approximately, both in a vertical and horizontal direction at this stage. Insure that the lever with the arrow on the titrator exchange unit is turned fully to the right. This is the normal position when the burette is full. Slide the titrator platform so that positioning pins on the base and slot on the exchange unit lever are aligned with guides on the Dosimat. (Note: the positioning pins on the base tell the Dosimat the burette size and the slot on the valve lever permits the system to open and close the valve). If the plunger and shaft on the dosimat are correctly aligned, the titrator assembly will click into place and the display on the instrument will read '00.00'. If it is incorrectly aligned the assembly will not ease in to place and you must remove it and adjust the positioning of the plunger shaft. (A hooked tool is provided to assist in this operation).

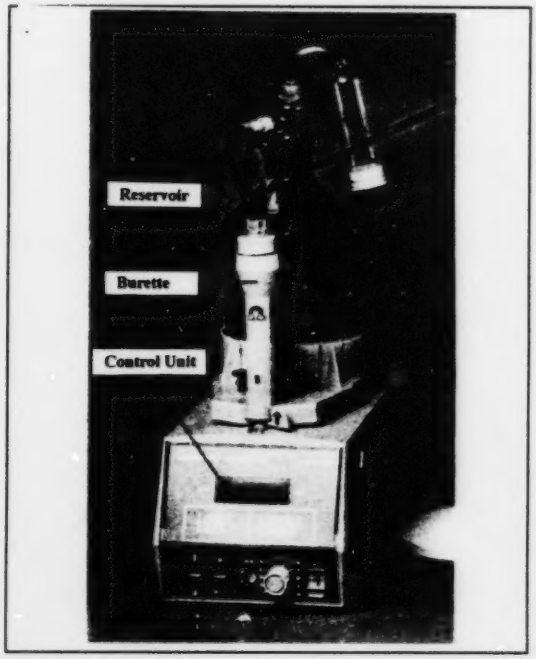

Figure 6. Dosimat and titrator assembly.

After you have successfully attached the 'snap-in burette unit', mount the titrant reservoir assembly and attach tubes linking it, the burette and the exchange unit as illustrated in Figure 6. (Note: It is important to ensure that the screw-thread fittings are tight to keep air from entering the system. Poor performance of the system can sometimes be traced to air in the lines.) One of the tubes is connected to the dispensing tip which is immersed in the sample during titration. A one-way valve prevents titrant leaking into the sample when there is no flow from the Dosimat. Mount the dispensing tip beside the probe from the colorimeter on the probe support. The tip should be as close as possible to the sampling region of the colorimeter probe.

Once you have completed these steps, purge air from the burette, valves and lines. The Dosimat offers several ways of doing this. The simplest involves setting the function switch found on the lower right to '5' and flipping the left hand toggle switch up. In this mode, the instrument automatically empties and refills the burette once. Do this several times and at the same time tap parts of the system to dislodge bubbles. The fill/refill procedure can also be accomplished by setting the function switch to '1' and lifting and holding up the toggle switch for emptying, and pressing down for refilling the burette.

When you are satisfied that there are NO more bubbles in the system, plug the two cables from the back of the computer into the 50 pin card edge connectors on the back of the Dosimat (Figure 7). The plugs are labelled and have a groove to indicate the correct orientation. The plugs must be fully in the sockets for the system to operate correctly. (Note: The Dosimat manual controls will not work while the computer cables are attached.)

Finally plug in magnetic stirrer and ready stirring bars.

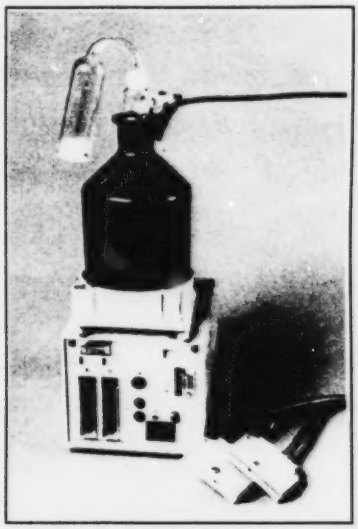

Figure 7. Back of Dosimat showing cable connections.

3.5 Set up of Oxygen Determination Program

Ensure that the diskette containing the program and database files is in the 'A' drive, and that the prompt A:\> is displayed. Place a new, formatted diskette in the 'B' drive. If you do not use a blank diskette you will not be able to open a database for your current round of samples. You can make a blank diskette by deleting all the files on a diskette or by formatting a new diskette. (Note: A new diskette is used at the beginning of the collection of each natural set of data, such as a survey or project. A single diskette can hold more data than you are ever likely to encounter in a typical field sampling program. Normally several diskettes with back-up are used on each cruise to prevent loss of data.) Instructions for formatting a diskette are found in Section 4.3.

To verify that the system is working, begin by typing 'OXYGEN' to start the program. The screen will appear as in Figure 8. Have several test samples ready, prepared following procedures outlined in Section 2.3. From the screen, use the down arrow key to highlight '(7) Perform Titrations'. Press <ENTER>. You will be presented with the following request from the computer:

'What is your sample identifier?'

Enter any number, preferably one you're not going to use on the cruise, and press <ENTER>. (Note: Letters, backspaces, and anything other than numbers will not be accepted by the system.) The system will then ask:

'What is your bottle number ? (9999 for none)'.

Enter 9999 and press <ENTER>.

A checklist will appear. Follow the instructions by placing a stirring bar in the first test sample, rinsing the colorimeter probe with distilled water and immersing it and the tip of the titrant dispenser in the sample. (Note: Adjust the magnetic stirrer to a speed at which the vortex does not draw bubbles into the sample, **not** as specified on the screen.)

Pressing <ENTER> will start the titration and display the % Transmittance and volume of titrant added on an X-Y graph. The sample colour will change from brown through yellow and be clear when the titration is complete. The computer will stop adding titrant when the curve reaches an inflection point at about 100% Transmittance, and will ask if the titration is complete. Type 'Y' for yes. The program will ask if you wish to do more samples. Do several test samples to verify that the system can produce a titration curve similar to that in Figure 9. If the system fails to produce such a curve, consult Section 4.7, Troubleshooting.

At this stage you may also set up your cruise database, run your standards and blanks or wait until you are prepared to start running samples. These operations are described in the following section. The Oxygen Determination program is described in more detail in Section 5.

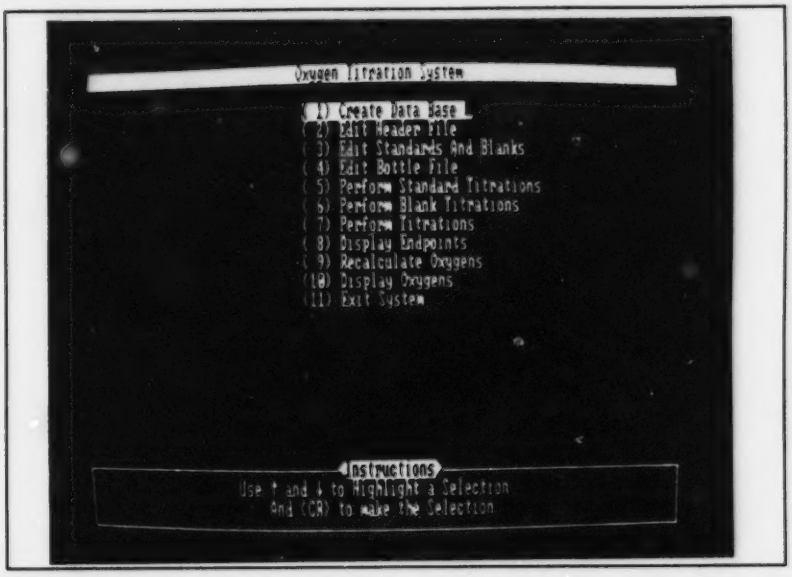
Oxygen Titration System
(1) Create Data Base
(2) Edit Header File
(3) Edit Standards And Blanks
(4) Edit Bottle File
(5) Perform Standard Titrations
(6) Perform Blank Titrations
(7) Perform Titrations
(8) Display Endpoints
(9) Recalculate Oxygens
(10) Display Oxygens
(11) Exit System
Instructions
Use ↑ and ↓ to Highlight a Selection
And (CR) to Make the Selection

Figure 8. Opening screen for oxygen titration system.

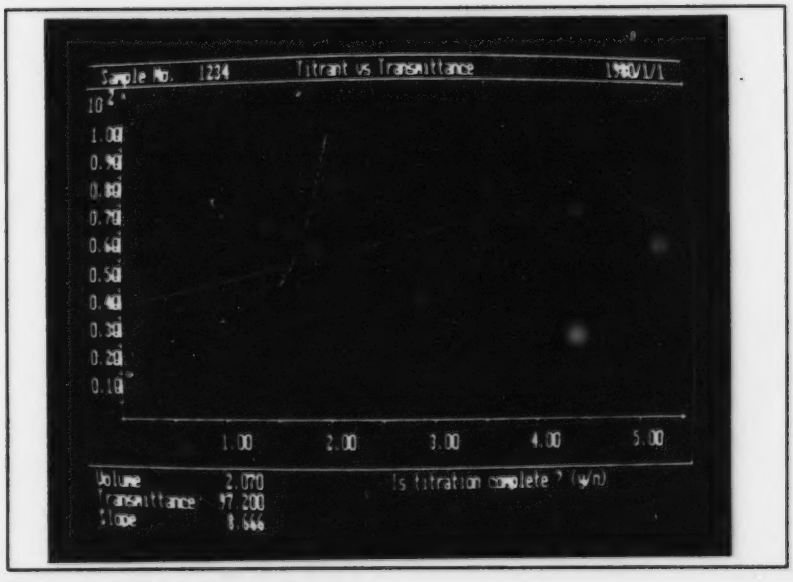

Figure 9. Sample titration curve.

4. OPERATING INSTRUCTIONS

4.1 Start-up Procedure

This section assumes that the operator has set up the equipment and has verified that it is performing properly. The following steps are carried out in day-to-day oxygen analysis.

Turn the colorimeter on for a warm-up of at least five minutes, longer in cold conditions. The Dosimeter and personal computer do not need a warming up period. During start-up of the computer, ensure that the printer is properly connected and on-line. An inadequate connection can lock-up parts of the oxygen determination program.

When turned on, the computer will 'boot' (load its operating system into memory and run). The program disk must be in the computer in the A drive in order to boot properly. The system will request current time and date. Fill in date (mm-dd-yy) and time (hh-mm-ss) as the system will place them on reports and data files as a time reference. Upon successful start-up the prompt A:\> is displayed.

Disconnect the Dosimat from the computer by unplugging the 'A' lead. Set at mode 5, and push the left toggle up. The Dosimat will automatically empty itself (a volume of 5 mL) and then refill. Tap the tubing during emptying to get bubbles out. Plug in the 'A' connector. The system is now ready to go. (Note: While the Dosimat is connected to the computer all its inherent functions are disabled. Disconnecting the 'A' lead breaks the connection with the computer enables the Dosimat functions.)

After the warm-up period, check the colorimeter adjustment by placing the probe in distilled water. No adjustment need be made as long as transmittance falls in the 95-100% range since the titration is based on transmittance differences, not absolute values. If necessary, adjust to approximately 100 % transmittance using the adjustment knobs on the front of the instrument and follow the procedure in Section 3 for setting 0 % Transmittance (Figure 10).

Every few days gently clean the exposed optical surfaces of the fibre optic probe (lens and mirror) with a cotton swab (Q-tip and water). Neither the probe nor the tube for dispensing titrant should be left immersed in a solution for prolonged periods to avoid degradation of the silver reflective coating on the mirror. The dispensing tip does not

appear to be affected, however, by long periods of immersion. The tip has a one-way valve which prevents titrant from leaking into the sample when not under pressure from the Dosimat.

Check and ensure that there is an adequate supply of printer paper.

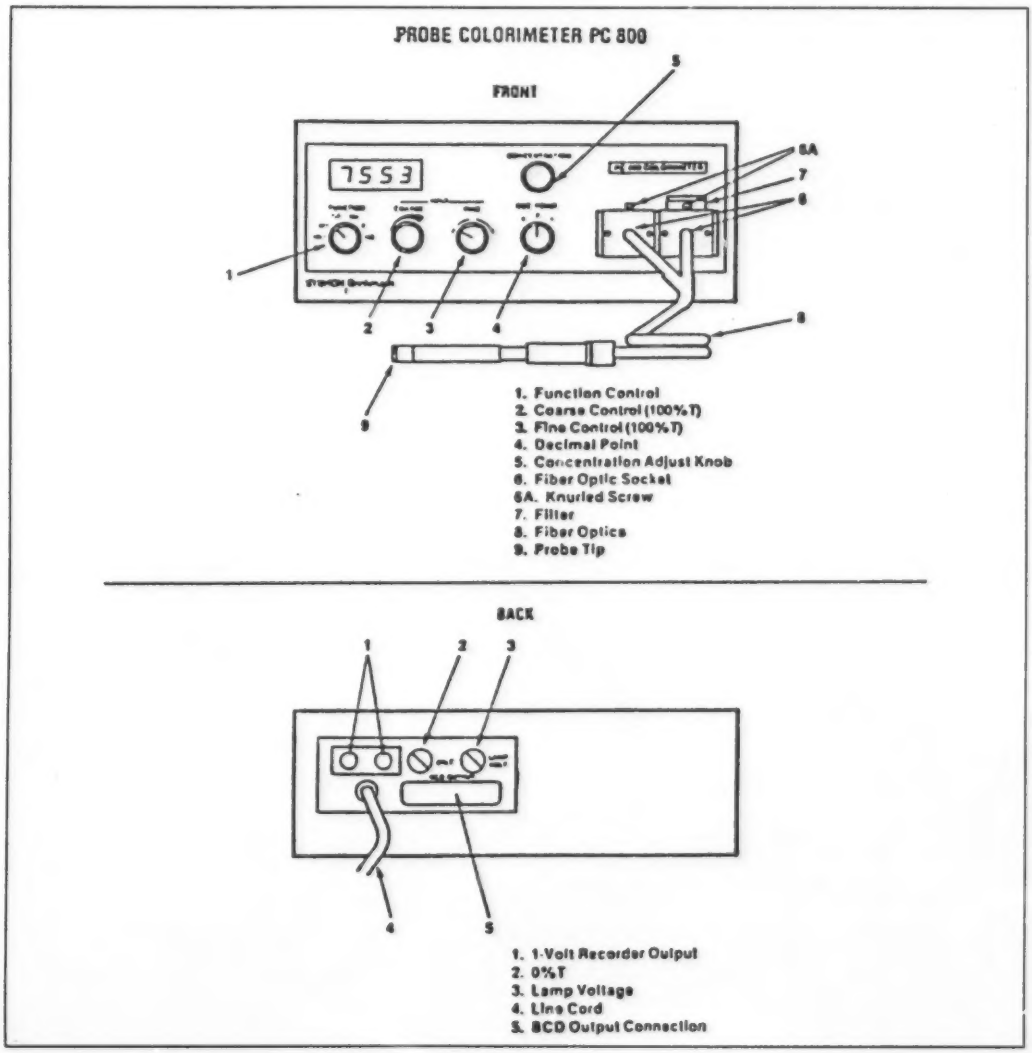

Figure 10. Schematic of colorimeter.

4.2 Titrating a Test Sample

Titrate a test sample prior to starting a sample run to ensure that the system is operating properly. Prepare test samples following procedures outlined in Section 2.3.

Type 'OXYGEN' to start the program. The screen will appear as in Figure 8 .
From the screen, use the down arrow key to highlight '(7) Perform Titrations'. Press <ENTER>. You will be presented with the following request from the computer:

'What is your sample identifier?'

Enter any number and press <ENTER>.

The system will then ask:

'What is your bottle number ? (9999 for none)'.

Enter 9999 and press <ENTER>.

A checklist will appear. Follow the instructions by placing a clean stirring bar in the sample, rinsing the colorimeter probe and the tip of the titrant dispenser with distilled water and immersing them in the sample. (Note: The speed setting instructions on the screen are not correct. Adjust the magnetic stirrer to a speed at which the vortex does not draw bubbles into the sample).

Pressing <ENTER> will start the titration and display the % Transmittance and volume of titrant added on an X-Y graph. The sample colour will change from brown through yellow and be clear when the titration is complete. The computer will stop adding titrant when the curve reaches an inflection point at about 100% Transmittance, and will ask if the titration is complete. Type 'Y' for yes. If the answer is no, the Dosimat will keep adding titrant until the endpoint is reached. The program will ask if you wish to do more samples. If the titration curve appears similar to Figure 9 you may begin analysing samples. If the system fails to give you a curve that ressembles Figure 9, consult Section 4.7, Troubleshooting.

4.3 Setting Up a Database

One of the features of the computer program is that it keeps track of all the data it needs to calculate and report oxygen concentration and stores it for later use. The data are stored on the diskette on the B: drive in several files, including a 'header' file, containing information identifying the cruise or sample run, chief scientist, date of analysis etc.; and several files containing information on volumes of sample bottles, standard and blank titrations, sample titration results, and oxygen determinations.

One complete set of files--the database--is stored on each floppy diskette. Normally a new set of files should be created on a new diskette each time the 'header' information changes. For instance, the 'header' file contains information on volumes of reagents added routinely to samples. That information can change in certain circumstances during a cruise or run of lab analyses. If so, set up a new database file.

Creating a database has two basic steps. First the program copies a model 'header' file and several working files to a blank diskette. The model 'header' file must then be edited to add information referring to the current group of samples.

To create a database you must have a blank, formatted diskette on which data and header information will be stored. Formatting is a procedure for organizing the memory on computer diskettes. If your diskette hasn't been formatted, do it now. It saves time if you format your diskettes in advance. (Note: A formatting program is present on the program diskette. To format a diskette, you must exit the OXYGEN Program, ensure that the program diskette is in the A: drive, and place an unformatted diskette is in the B drive. At the prompt A:\> type FORMAT B:. The system will tell you to insert the diskette in the B drive. Press <ENTER> and wait for the disk to be formatted. You will then have the option of either formatting another disk or quitting.)

To create the database, run the program by typing 'OXYGEN followed by <ENTER>. The first system screen will be displayed. Select the 'Create Data Base' option by using the down key and pressing <ENTER>. You will see the message:

> Insert a BLANK FORMATTED diskette in drive B:
> Hit any key to continue

In order to prevent you from erasing or writing over data you have already collected, the system will not set up a database on a diskette containing files. The system will display the message:

Disc in drive B is not blank. You must use a FORMATTED DS DD diskette that contains NO files.
Hit any key to continue

When you have correctly inserted a blank formatted diskette, press <ENTER>. The program will display the messages:

Copying: Main Header from A: to B:
Copying: Standards and Blanks from A: to B:
Copying: Bottle File from A: to B:
Hit any key to continue

Now that you have created the database, you must edit the header information to replace it with information relating to your current round of samples. Select '(2) Edit Header File' and press <ENTER>. The screen will show a form on which information relating to the sampling is displayed. The spaces will already be filled with information which you must change to correctly represent the current situation. Selections A to G and Q are for optional identifier information and you need only fill in enough for your purposes. Selections H to K give information particular to the oxygen titration procedure and should correspond to parameters you are using. Selections L to O are for the statistical routine which monitors the titration and should not be changed. P ('Printer Attached') must be filled out with either 'Yes' or 'No'.

To change a selection, type the corresponding letter and press <ENTER>. Then type the information and press <ENTER> again. Modify the remaining information on the form and then press 'X' and <ENTER> to return to the first screen. You are now ready to begin titrating.

4.4 Standards and Blanks

The oxygen determination procedure requires that several standards be titrated to allow adjustment for changes in the strength of the sodium thiosulphate solution used as titrant. Blanks give a correction for the influence of components of the reagents on the oxygen measurement.

Standards--The database may already contain previous entries for standards and blanks. These must be removed before you begin titrating standards or blanks. If at this point you select '(5) Perform Standard Titrations' or '(6) Perform Blank Titrations' and press <ENTER>, you may get the message:

ERROR : Maximum number of STANDARDS (BLANKS) has been reached !

This is because standards and blanks (five each) have not been deleted from the standards and blanks file. After displaying this error message, the program will return you to the first screen and you will proceed to select '(3) Edit Standards and Blanks'

From the first screen, select '(3) Edit Standards and Blanks' and press <ENTER>. The following screen will appear:

Edit Standards and Blanks

(1) Display Standards and Blanks
(2) Modify Standards
(3) Delete Standards
(4) Modify Blanks
(5) Delete Blanks
(6) Return to Main Menu

Select '(3) Delete Standards' and press <ENTER>. Up to five standards and the corresponding titration volumes will be displayed with the comment

Standard number to DELETE or (Q)uit ?

Type the numbers one at a time and press <ENTER>. When all the standards have been deleted type 'Q' to return to the '(3) Edit Standards and Blanks' screen menu. At this point, follow a similar procedure to delete the values for reagent blanks contained in the database. This will have to be done for your new set of blank titrations.

After you have deleted old standards and blanks, select '(6) Return to Main Menu' and press <ENTER> to return to the first screen. Every time you return to the main menu, the program writes the standards and blanks file to the diskettes in both A: and B: drives. The following message is displayed:

Updating Standards and Blanks file B:STNDBLNK.DTA
Duplicating on drive A: as file A:stndblnk.dta for use on subsequent runs

Now select '(5) Perform Standard Titrations' and press <ENTER>. The following instructions will be displayed on the screen:

1. Ensure DOSIMAT, COLORIMETER and MAGNETIC STIRRER are ON.
2. Rinse COLORIMETER PROBE with DISTILLED WATER.
3. Place FLASK on STIRRER, insert MAGNET and COLORIMETER PROBE making sure PROBE SLOT is covered.
4. Place MAGNETIC STIRRER on #4 or #5
5. Hit RETURN when ready

(Note: Adjust the magnetic stirrer to a speed at which the vortex does not draw bubbles into the sample, **not** as specified on the screen.)

Prepare standards by pipetting 10 mL of 0.01 N potassium iodate (KIO3) standard into a clean sample bottle, filling the bottle with distilled water, and adding the sulphuric acid and sodium iodide-sodium hydroxide (alkaline azide) reagents. A drop of wetting agent is added to each sample to reduce the tendency of bubbles to stick to the surfaces of the optical probe. Preparation of standards has been outlined in detail in Section 2.

Pressing <ENTER> will start the titration and display the % Transmittance and volume of titrant added on an X-Y graph. The sample colour will change from brown through yellow and be clear when the titration is complete. The computer will stop adding titrant when the curve reaches an inflection point at about 100% Transmittance, and will ask if the titration is complete. If for some reason the sample still has a yellow hue, type 'N' (for no) <ENTER> and the titration will continue briefly. (Note: Sometimes bubbles in the sample can interfere with the determination of the endpoint. The titration can be completed satisfactorily by choosing the 'N' option at this point.) The titration curve should ressemble Figure 9. If the system fails to give you a satisfactory curve, consult Section 4.7, Troubleshooting.

The program will ask if you wish to do more standards. Press 'Y' for yes and repeat the procedure. Do at least 3 standards (the system will accommodate up to five standards and blanks) and accept those within 0.02 mL of each other. Delete outlying values if more are analyzed, since the system uses all the values available to it in the database in computing average values and ultimately oxygen concentrations. To view your standards, choose the '(1) Display Standards and Blanks' option from the '(3) Edit Standards and Blanks' screen.

Blanks--Return to the starting screen menu. The database at startup contains values for blanks from previous sample runs. If you have not already removed these values, do so now. If at this point you select '(6) Perform Blank Titrations' and press <ENTER>, you may get the message:

ERROR : Maximum number of BLANKS has been reached !

This is because the maximum of five standards and blanks are in the standards and blanks file. After displaying this error message, the program will return you to the first screen and you will proceed to select '(3) Edit Standards and Blanks'

From the first screen, select '(3) Edit Standards and Blanks' and press <ENTER>. The following screen will appear:

Edit Standards and Blanks

(1) Display Standards and Blanks
(2) Modify Standards
(3) Delete Standards
(4) Modify Blanks
(5) Delete Blanks
(6) Return to Main Menu

Select '(3) Delete Blanks' and press <ENTER>. Up to five blanks and the corresponding titration volumes will be displayed with the comment

Standard number to DELETE or (Q)uit ?

Type the numbers one at a time and press <ENTER>. When all the blanks have been deleted type 'Q' or select '(6) Return to Main Menu' to return to the first screen.

Now select '(5) Perform Blank Titrations' and press <ENTER>. The following instructions, which are identical to those for titrating standards, will be displayed:

1. Ensure DOSIMAT, COLORIMETER and MAGNETIC STIRRER are ON.
2. Rinse COLORIMETER PROBE with DISTILLED WATER.
3. Place FLASK on STIRRER, insert MAGNET and COLORIMETER PROBE making sure PROBE SLOT is covered.
4. Place MAGNETIC STIRRER on #4 or #5
5. Hit RETURN when ready

Prepare reagent blanks by pipetting 1 mL of 0.01N potassium iodate solution into a clean sample bottle, filling the bottle with distilled water, and adding 1.5 mL sulphuric acid and alkaline iodide reagent. Following mixing, add the manganous chloride and a drop of wetting agent.

Follow the instructions displayed on the computer screen with the exception that the magnetic stirrer need only be adjusted to create enough of a vortex to mix the sample but not to draw bubbles into the sample flask.

When you press <ENTER> the system will begin titrating the first blank. A titration curve will be displayed on the screen. The program will determine the endpoint, stop the titration and ask if the titration is complete. If for some reason the sample still has a yellow hue, type 'N' (for no) <ENTER> and the titration will continue briefly. (Note: Sometimes bubbles in the sample can interfere with the determination of the endpoint. The titration can be completed satisfactorily by choosing the 'N' option at this point.) The titration curve should ressemble Figure 9. If the system fails to give you a satisfactory curve, consult Section 4.7, Troubleshooting.

Blank titrations have two steps. After the first titration is complete, a second aliquot of potassium iodate must be titrated to determine the blank. Consequently, following the first titration, the system sets up immediately to begin another titration without asking if you wish to titrate a new sample.

Pipette a second millilitre of potassium iodate into the sample and press <ENTER>. The system will repeat the steps above. This time, however, after pressing 'Y' to indicate the titration is complete, the system will ask if you wish to carry out a further blank titration. (Note: The reagent blank is the difference between the first and second titration values. Preparation of reagent blanks is outlined in detail in Section 2.3).

Repeat the procedure until a satisfactory level of precision is obtained between two blank determinations. To view standards and blanks, choose the '(1) Display Standards and Blanks' option from the '(3) Edit Standards and Blanks' screen. Remove any outlying values using the '(5) Delete Blanks' option. The program uses all the values in the Standards and Blanks file to calculate a blank value.

Having determined standards and blanks, you are now ready to titrate samples.

4.5 Analyzing Samples

Ensure that you are positioned at the opening screen menu and have your samples ready. Use the down arrow key to highlight '(7) Perform Titrations'. Press <ENTER>. The system will print time, date, and headings for a data table, and will display the following request:

What is your sample identifier?

Enter a number which uniquely identifies the sample and press <ENTER>. (Note: If the number has already been used, the system will ask if the this sample is a replicate. The system accepts replicates, so if you indicate 'Y' to this question, the system will continue to the next step. If you answer 'N', the system will repeat the above request for a sample identifier.)

The system will then ask:

'What is your bottle number ? (9999 for none)'.

Sample bottles do not have identical volumes and assigning a bottle number enables logging of a calibrated volume for each. Occasionally a bottle may be used which does not have a number or for which the stopper has been switched with another bottle. Use a valid sample number and a bottle number of '9999' and press <ENTER>. This logs the data, so that later, when you obtain a volume, you can calculate the oxygen concentration manually. (Note: If you enter the wrong bottle or sample number, note both the correct and incorrect values. Later, correct the sample data file using a text editing program, and use the recalculate option to generate new oxygen concentrations.)

The following checklist will appear:

1. Ensure DOSIMAT, COLORIMETER and MAGNETIC STIRRER are ON.
2. Rinse COLORIMETER PROBE with DISTILLED WATER.
3. Place FLASK on STIRRER, insert MAGNET and COLORIMETER PROBE making sure PROBE SLOT is covered.
4. Place MAGNETIC STIRRER on #4 or #5
5. Hit RETURN when ready

Follow the instructions by placing a clean stirring bar in the sample, rinsing the colorimeter probe with distilled water and immersing it and the tip of the titrant dispenser in the sample.

Pressing <ENTER> will start the titration and display the % Transmittance and volume of titrant added on an X-Y graph. The sample colour will change from brown through yellow and be clear when the titration is complete. The computer will stop adding titrant when the curve reaches an inflection point at about 100% Transmittance, and will ask if the titration is complete. If there is any yellow coloration to the sample, type 'N' for NO and press <ENTER>. The system will continue titration and will ask again if the titration is complete. When it is, type 'Y' for yes and press <ENTER>. The program will ask if you wish to do more samples. Continue doing all the samples in the lot. Check regularly that the system is producing titration curves similar to that in Figure 11. If the system fails to give you a satisfactory curve, consult Section 4.7, Troubleshooting, before conducting further analyses.

Titrations normally take 1.5 to 2 minutes. Each time a sample is titrated successfully the program logs the data in the B: drive and prints the titrant volume, oxygen concentration, bottle number and sample identifier. At the end of the sample run, the printer output provides a record of the analyses.

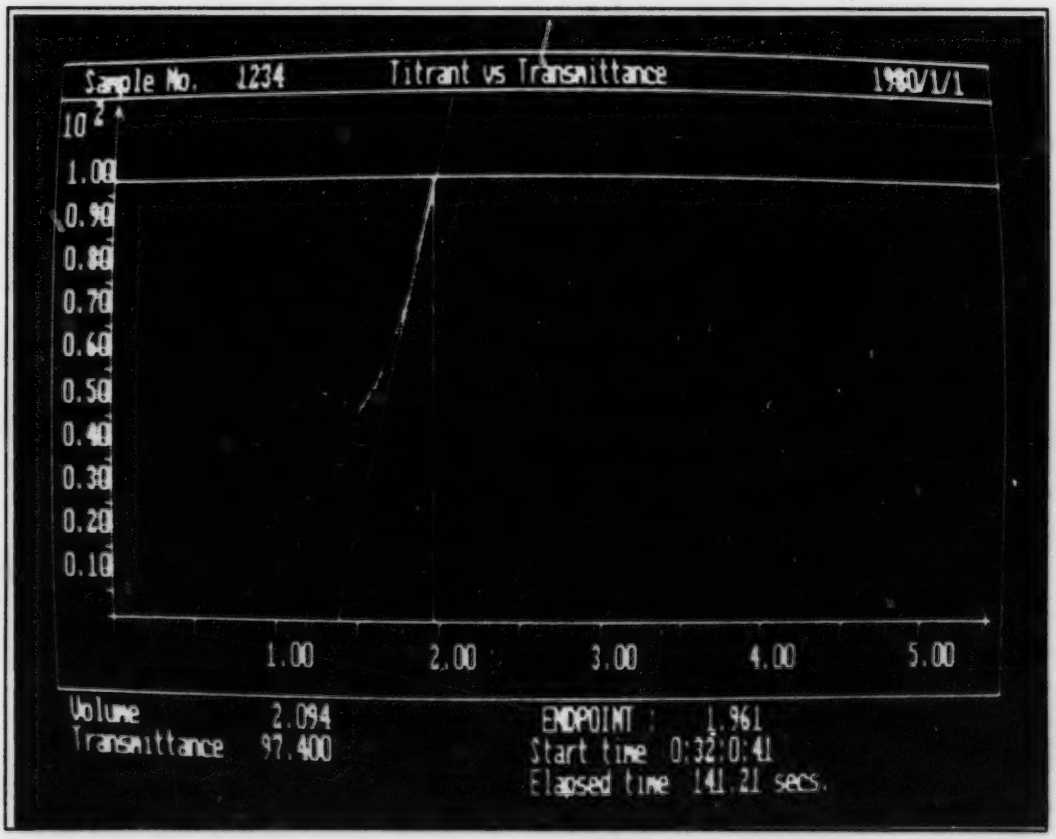

Figure 11. Titration curve display and end point calculation from 'Oxygen' program.

4.6 Data Management

Under normal circumstances, the OXYGEN program looks after data management for you. The program stores data on the diskette on the B: drive in several files, and prints the oxygen concentrations of samples as they are titrated. In addition, you can view or print out the stored data at any time, and can even recalculate oxygen measurements.

Several options on the first screen menu allow you to view and edit data:

- **'(2) Edit Header File'** displays information identifying cruise and sample run reagent volumes, statistical criteria for determining endpoint for the titration, and information on the printer status. Normally you do not have to edit this file during the cruise or run. Information is entered in it at the beginning when you set up a new database diskette (See Section 4.3). Changes made to reagent volumes and titration parameters will be used in subsequent oxygen determinations.

- **'(3) Edit Standards and Blanks'** allows you to view and edit standards and blanks. A screen menu lists various options.

- **'(4) Edit Bottle File'** displays the calibrated volumes of oxygen bottles. A screen menu displays options to list, add, modify, or delete bottles from the file, or print the list of sample bottles and calibrated volumes.

- **'(8) Display Endpoints'** displays the volume of titrant used for each oxygen determination.

- **'(10) Display Oxygens'** displays oxygen concentration in samples.

If a parameter used in determining oxygen concentration is incorrect in the 'header file' it results in an incorrect determination of oxygen concentration. For example dispensers used in dispensing reagent sometimes deliver the wrong volume because of bubbles, plunger sticking, etc. The system allows you to correct the parameter in the header file and then recalculate the oxygen values using option '(9) Recalculate Oxygens'. This option uses the titrant volumes previously determined for the samples and uses the new values in the header file for the calculation. (Note: Be careful when using this option since it also arranges the samples in numerical order and affects all the samples in the file). To be safe make a backup copy of the diskette in the B: drive before using the recalculate option. For more information on the recalculate option consult Section 5.

4.7 Troubleshooting

A user may occasionally encounter problems with the operation of the system. Some of the solutions are outlined below:

Instrument	Problem	Solution
Colorimeter	Transmittance cannot be adjusted above 90%.	Lamp voltage may be low. Adjust to between 7.5 and 8 Volts.
		Connections of light guides with the colorimeter may be loose.
		Fibre optic sensor may be dirty.
Titrator	Front controls don't function.	The titrator is disabled while connected to the computer. Breaking the connection with the computer will let you use the titrator controls.
	Questionable titrant volumes.	Tubing connections on titrator may be loose and the system is sucking air.
Oxygen Determination Program	Titration not completed.	Small bubbles on the sensor probe; add wetting agent.
	Titration takes a long time.	Colorimeter is not adjusted properly. Transmittance in pure water should be 90-100% and instrument should be zeroed.
	Titration repeats without asking for new sample information.	You have used the 'Perform Blank Titrations' routine instead of 'Perform Titrations'.
	Program 'locks'.	Check connections with titrator, colorimeter and printer. Reboot the computer.
	System shows a runtime error.	Check all connections with titrator, colorimeter and printer. Rerun OXYGEN program.

5. SYSTEM IN DETAIL

5.1 Computer

The oxygen titration system uses an IBM PC or compatible computer in which a Data Translation, DT2806 multi-function output subsystem host board and three Data Translation DTX350 digital input/output modules, have been installed. The three modules plug into the host board which is mounted in one of the computer's 62-pin expansion slots. Each module has an iSBX J1 connector which connects it to the main board and a card edge connector for interfacing with the cables.

The DT2806 host board has three multi-function timer/counters corresponding to each of the digital input/output modules. With the three modules in place the board can handle 72 channels of digital input/output, 24 channels of digital/analog signals, or 80 single ended analog/digital channels.

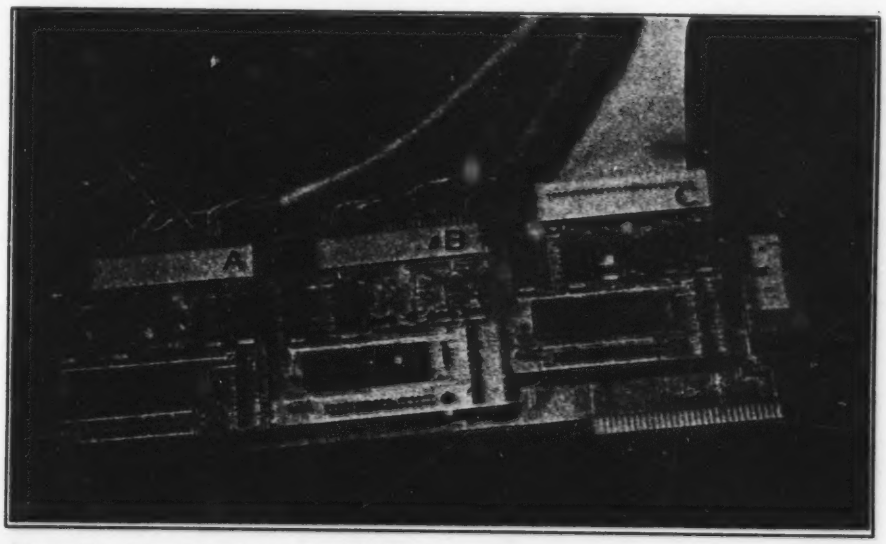

Figure 12. Expansion board for IBM PC computer.

The digital I/O modules labelled A and B in Figure 12 are connected by cables to the Dosimat and module C is connected to the colorimeter. Wiring of each connection will be described with the corresponding instrument. Note that the iSBX J1 connector which links the digital I/O modules with the cables, is not industry standard and the wires in the

cable to which the colorimeter and Automatic Titrator are connected must be offset by one. Thus, wire one in the cable should be connected to pin two on the iSBX connector on the modules and so on.

Jumper pins on each of the modules are installed as indicated in Figure 13.

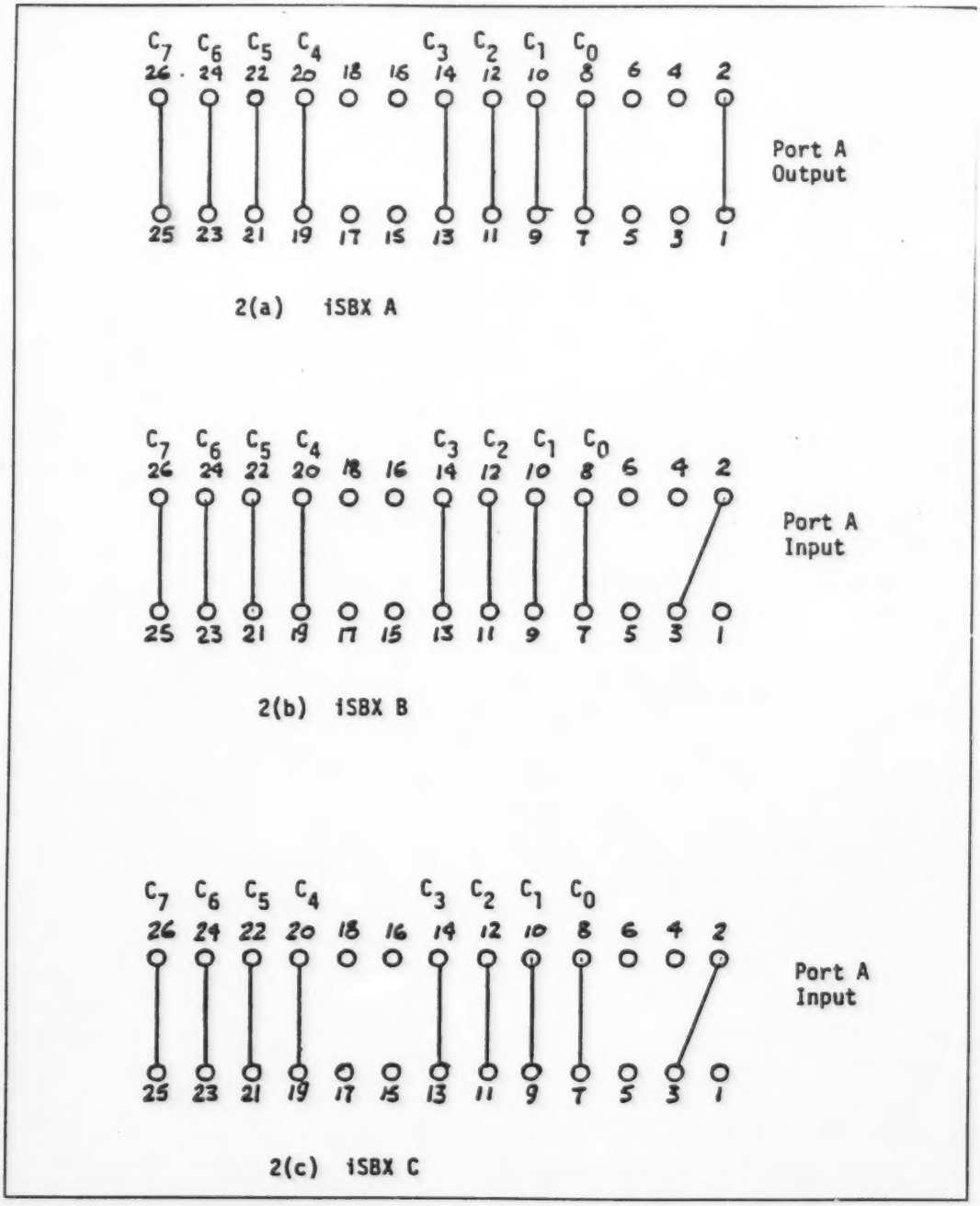

Figure 13. iSBX Jumper configuration.

5.2 Automatic Titrator -- Metrohm 655 Multi-Dosimat

Program Control--The Metrohm 655 Multi-Dosimat is a precision motor-driven piston burette which has a self-contained microprocessor to allow automatic operation. The system can be operated manually or entirely under the control of a computer. The Dosimat has eight modes but only two are used by the oxygen titration program:

Mode 1: Dosing continues as long as the dosing command is active. The speed of fluid flow is variable and controllable.

Mode 2: On command, 1/10,000 of the 5 mL burette volume is expelled.

An explanation of the other modes is contained in the manual for the instrument. Mode 5 is useful in manually purging air from the burette and supply lines when the system is first set up. This mode allows the burette to empty until the upper volume limit of the system (in this case 5 mL) has been reached).

At the start of a titration the oxygen titration system places the Dosimat in Mode 1 and the titration begins. The program can instruct the Dosimat to dispense the titrant at four flow rates, ranging from slow to fast, depending on the transmittance of the solution. Above a transmittance of 75 % the titrant is dispensed at the slowest rate. The system can also command the Dosimat to provide minute pulses of titrant, as when the operator requests to continue the titration after the system has determined endpoint. In this case the system sets the Dosimat to Mode 2 which gives pulsed addition of as little as 1×10^{-4} of the burette volume (5×10^{-4} mL for a 5 mL burette).

The Dosimat has two ports at the rear, each with a 50-pin edge card. Newer versions of the instrument have more advanced capabilities, which will not be discussed here nor can be accommodated by the current system. Port A is used for computer control of the instrument and Port B is used for reading the volume of titrant dispensed, measured and transmitted by the instrument in Binary Coded Decimal (BCD) format.

While the Dosimat is connected to the computer, its internal functions are disabled. To carry out functions such as flushing the system, disconnect the computer cables from the unit and then use manual operation.

The Dosimat is designed to handle burettes having different volumes but the oxygen determination program reads the actual volume titrated from the instrument, so it should be possible to use the program with different burettes.

5.3 Brinkman PC/8000 Colorimeter

The colorimeter measures transmittance and absorbance using a fibre optic probe immersed in the solutions to be tested. It can also be set up to indicate concentration. For the purposes of the oxygen titration system, only the first capability is required. The system consists of:

- Console;
- Fibre optic light guide and probe; and
- Connector cable for computer.

Warmup--The colorimeter should be turned on at least 5 minutes before use and longer in cold conditions.

Adjustments--The colorimeter should be checked regularly for proper adjustment. After the instrument is warmed up, check that the lamp voltage falls in the optimal range of 7.5 to 8 volts and adjust if necessary. To change lamp voltage, set the left hand knob to LAMP VOLTAGE and use the slotted adjustment knob on the back of the instrument. Adjust the zero transmittance by disconnecting the right hand light guide from where it plugs into the instrument and adjusting to zero using the slotted knob on the back of the instrument. After re-connecting the light guide, check the 100% transmittance setting by immersing the probe in distilled water. The sampling region in the probe must be completely submerged and free of bubbles for correct adjustment. Use the COARSE and FINE adjustment knobs on the front of the colorimeter to make adjustments. If you cannot adjust transmittance to 100 %, increase the lamp voltage and try again.

Fibre Optic Probe--The stainless steel fibre optic probe connects via a light guide to the front of the instrument. The sampling region of the probe is a 1 cm long opening which, with the built-in mirror, forms a 2 cm path through

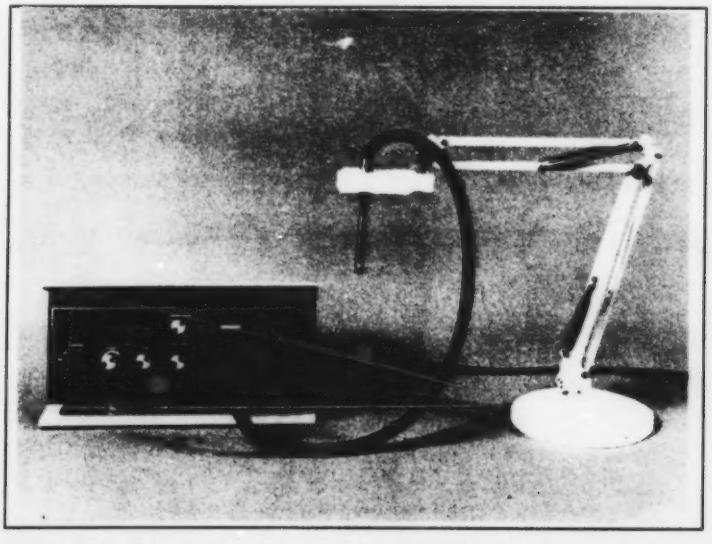

Figure 14. Colorimeter, fibre optic probe and articulated arm.

which light from the end of the incoming fibre optic light guide can pass. Light from the instrument passes through the sampling region and returns to the instrument which measures changes due to absorption in the sample.

Although the probe is durable, it should not be immersed in fluids for long periods when not in use. The probe should be cleaned occasionally with soft tissue and blotted dry if possible. In normal use, it should be rinsed with distilled water between samples.

The fibre optic probe may be conveniently supported by inserting it, and the dispensing tip of the Dosimat, through holes bored in a block of Teflon. The block is attached to the end of the articulated arm. This arrangement has been used routinely on Bedford Institute of Oceanography cruises (Figure 14).

Outputs--The colorimeter supplies a readout of the display in Binary Coded Decimal (BCD) through a 50-pin Centronics type connector at the back of the instrument. A cable running from the colorimeter is connected to the digital I/O module C on the expansion board in the computer (Figure 12). Note that the iSBX connector on the digital I/O module is not industry standard and that the cable connections must be advanced by one to the connector. Thus cable 1 would be connected with pin 2 on the iSBX connector.

The colorimeter also has outputs for a chart recorder. These deliver up to 1 Volt of current proportional to transmittance, with 1 Volt equivalent to 100% transmittance.

5.4 Oxygen Titration Software (OXYGEN)

Control of Instrumentation

The computer program called OXYGEN configures the computer for input or output from the devices in the system, controls dispensing and refilling of the Dosimat, reads values from the colorimeter and Dosimat, determines endpoint, and provides an interface for the user with various screens (menus) containing options for running the system and displaying and editing data. The source code in the PASCAL programming language is available from: Fisheries and Oceans Canada, Physical and Chemical Sciences Branch, Bedford Institute of Oceanography, P.O. Box 1006, Dartmouth, Nova Scotia B2Y 4A2.

Determination of Endpoint

The program determines endpoint by a statistical analysis of the plot of transmittance versus titrant volume. During a titration, the system monitors transmittance and titrant volume, and logs them to the temporary file 'RAW.DTA' on the B: drive. Concurrently, it removes spikes and spurious points from the data using a filter which selects the 'median' from among a 'window' containing a number of points, usually seven, specified in the header file. The filtered data are logged to the 'REDUCED.DTA' file on the B: drive and are used to compute the slope of the titration curve. The system keeps track of the maximum slope in a given titration and compares each slope with that value, resetting the maximum if necessary. It also runs a check for possible endpoint by checking if slope has fallen to a specified percentage of the maximum slope (10 % or less). The cutoff percentage can be set by the user as "Slope Tolerance' in the header file.

Values for transmittance, titrant volume, and slope are displayed and successive points in the titration curve are plotted on the screen. If the system determines that the endpoint has been reached, (i.e. slope of 10% or less of maximum) it stops the addition of titrant and prompts the user for an OK to quit the titration.

To compute endpoint, the program calculates a linear regression relationship for the steep part of the curve and one for the asymptotic region and then determines the x-intercept of the point at which the two lines intersect. The number of points used in determining the separate regressions are different and are specified in the header file ('Number of points to fit' refers to the regression for the steep part of the curve; and 'No. of Points in Baseline' refers to the asymptotic portion). For the steep part of the curve, the range of points selected is centred on the point of maximum slope. The endpoint, or x-coordinate of the point where the lines intersect, is simply:

$$\frac{b_2-b_1}{m_1-m_2}$$

where b_1 and b_2 are the y-intercepts of the curves and m_1 and m_2 are the slopes.

In addition to computing and displaying the endpoint, the program plots the two regressions and draws a line from the intersection to the x-axis. It computes oxygen concentration, and logs the titrant volume, sample number, and computed oxygen concentration in a file on the B: drive. The raw and reduced data remain in a file on the B: drive until a new titration is begun. Thus a titration may be reconstructed from the

temporary data if the procedure is interrupted after the endpoint has been determined but before a new sample has been titrated.

User Interface

The program user interface consists of a number of screens (menus) which list various options offered by the system. The user highlights the selection using the <UP> and <DOWN> keys on the computer numeric keypad and finalizes the selection by pressing <ENTER>. Once an operation has been selected, the system may prompt the user for information, display instructions, or display relevant data.

Main Screen Menu

Typing OXYGEN <ENTER> at the A:\> prompt starts the program. The main screen menu is displayed with the following options:

Oxygen Titration System

(1) Create Data Base
(2) Edit Header File
(3) Edit Standards and Blanks
(4) Edit Bottle File
(5) Perform Standard Titrations
(6) Perform Blank Titrations
(7) Perform Titrations
(8) Display Endpoints
(9) Recalculate Oxygens
(10) Display Oxygens
(11) Exit system

The various options are as follows:

(1) *Create Data Base*--This option sets up a new database by copying the following files from the A: to B: drive:

HEADER.DTA	-cruise information and titration parameters
BOTTLE.DTA	-bottle volumes
STNDBLNK.DTA	-endpoints for standards and blanks

The system requires that a blank, formatted diskette be in the B: drive. The DOS

FORMAT utility is stored on the disk with the oxygen program to enable easy formatting of diskettes. The following files in addition to those listed above are also on the program diskette:

Drivers:

ATT.BGI
CGA.BGI
HERC.BGI
PC3270.BGI
EGAVGA.BGI
IBM8514.BGI

Utilities:

COMMAND.COM
FORMAT.COM
PRINT.COM
DISKCOPY.COM

System Data files:

VBOT.DTA
STNDBLNK.DTA
BOTTLE.DTA
HEADER.DTA

Program files:

MAKEBOT.EXE
MAKHEAD.EXE
OXYGEN.EXE

In addition, the following two files containing colorimeter outputs are placed on the B: drive during titration:

REDUCED.DTA	(system management file)
RAW.DTA	(system management file)

(2) *Edit Header File*--This option allows the header file copied to the B: drive to be filled with information relating to the current cruise or run. The screen displays a form with the following information:

(A) Cruise Number
(B) Ship's Name
(C) Chief Scientist
(D) Cruise Date
(E) Analysis Location
(F) Analyst
(G) Analysis date
(H) Volume of KIO3
(I) Normality of KIO3
(J) Volume of Alkaline Iodide
(K) Volume of Manganous Chloride
(L) No. of Points to Fit
(M) No. of Points in Baseline
(N) Slope Tolerance
(O) Median Filter Window Size
(P) Printer Attached
(Q) Main Header Comments:

(A) through (G) describe the current cruise or analysis run.

(H) through (K) are standard parameters relating to the titration and are changed as needed.

(L) is the total number of points used in fitting a linear regression to the steep portion of the titration curve.

(M) is the total number of points used in fitting a linear regression to the asymptotic or level portion of the titration curve.

(N) is the percentage of maximum slope at which the program determines that an end point has been reached.

(O) is the number of points used by the 'median filter' to remove sporadic high points or spikes from the data.

(P) indicates if the printer is attached. (Note: The system will ask for a y/n response. If the printer is not connected or is off line, having '(P)' set to 'YES' will lock-up the system. The system will perform all functions except printing when '(P)' is set to 'NO'.)

(Q) takes three, 40-character lines of comments.

After the header file has been updated, typing 'X' will return the user to the main screen.

(3) *Edit Standards and Blanks*--This option provides access to the data file containing the results of standard and blank titrations. As the system computes an average of all standards and blanks in the file, all old standard and blank values must be deleted using

this option before new standards and blanks are determined. The system will hold up to five standard and five blank values. The options '(5) Perform Standard Titrations' and '(6) Perform Blank Titrations' will give the following error message if the maximun of five standards (blanks) have not been removed from the data file:

ERROR: Maximum number of STANDARDS (BLANKS) has been reached!

This message will not appear if there are fewer than five standards or blanks in the files.

The '(3) Edit Standards and Blanks' option displays the following menu:

Edit Standards and Blanks

(1) Display Standards and Blanks
(2) Modify Standards
(3) Delete Standards
(4) Modify Blanks
(5) Delete Blanks
(6) Return to Main Menu

'(1) Display Standards and Blanks' presents the following list of standard and blank values (expressed as mL of titrant):

Standard titration volumes

Standard # 1 1.991
Standard # 2 1.988
Standard # 3 etc.

and Blank titration volumes

Blank # 1 0.010
Blank # 2 -0.016
Blank # 3 etc.

Options from '(2) Modify Standards' to '(5) Delete Blanks' present the list of standards (blanks) as requested and the comment:

Standard (blank) number to DELETE (MODIFY) or (Q)uit ?

(Note: The system presents a new list after each DELETE (MODIFY) operation and repeats the comment, until 'Q' is pressed. This returns the 'Edit Standards and Blanks' screen menu. After the '(6) Return to Main Menu' option has been selected and <ENTER> has been pressed, the system writes the new values to both A: and B: drives, and displays the message:

> Updating Standards and Blanks file B:STNDBLNK.DTA
> Duplicating on drive A: as file A:STNDBLNK.DTA for use on subsequent runs.

The system copies these data files automatically whether or not changes have been made to the standards or blanks.

(4) *Edit Bottle File*--The bottle file contains a listing of the calibrated volume for each oxygen bottle in use in the particular institution or sampling program. Up to 999 oxygen bottles can be recorded. The file is stored on the program diskette and volumes of individual oxygen bottles are retrieved by the system during sample titrations for use in computing oxygen concentration. Normally the file is modified only when bottles are calibrated or when bottles are removed from circulation through breakage or wear. The bottle file can be changed at any time during a titration session.

This option displays the following selections:

> Edit Bottle File
>
> (1) List Bottles
> (2) Add Bottles
> (3) Modify Bottles
> (4) Delete Bottles
> (5) Print Bottles
> (6) Return to Main Menu

'(1) List Bottles' displays a numerically ordered list of bottle numbers and calibrated volumes. One screen at a time is displayed. Pressing <ENTER> will display the next screen. At the end of the list, the system prompts the user to press <ENTER> to return to the main screen.

Options 2 to 4 allow the bottle list to be changed. '(2) Add Bottles' displays the following request:

Enter bottle number and volume; Hit <CR> to end
--->

Type the bottle number (1-999) and the volume to two decimal places. The bottle number and volume in millilitres must be separated by a space. If you make an error while entering the values and correct it using the backspace key, the system will read backspace as a character and will display the message:

ERROR: Illegal input, non-numeric character!

and will again prompt for input of bottle number and volume. Then simply type in the correct value and press <ENTER>. If a bad value is entered into the system, the only way to change it is to leave the '(2) Add Bottles' option and select the '(3) Modify Bottles' option.

In the 'Add Bottles' option you cannot overwrite data for an existing bottle of the same number. If another bottle on the system has the same number, the following error message will be displayed:

ERROR: Bottle 870 already exists!

If the other bottle has been lost or discarded, then use the '(4) Delete Bottles' option first, before attempting to add the new bottle to the file.

The '(3) Modify Bottles' selection displays the prompt:

Enter bottle number and new volume; Hit <CR> to end.
--->

Enter the number and volume in millilitres to two decimal places separated by a space. The system will display the following error message if the bottle to modify is not found:

ERROR: Bottle 920 not found!

The '(4) Delete Bottles' selection displays the prompt:

Enter bottle # to DELETE; Hit <CR> to end.
--->

If you have entered the number of a bottle which is not stored on the system, the following error message will appear:

ERROR: Bottle 994 not found!

Ensure that you have entered the correct number to avoid deleting a valid bottle. If the number is in error, type 'backspace' and then <ENTER>. The system will indicate an error and will again prompt for the bottle number to be deleted.

The '(5) Print Bottles' option instructs the program to print a list of bottles and calibrated volumes. The system will give the message:

Printing list of Bottles.

If the header file indicates that no printer is attached, the system will give the error message:

ERROR: Printer exists flag set to NO in header file!

If the printer is not attached or off line, the message 'Printing list of Bottles' will still be displayed to the screen.

(5) *Perform Standard Titrations*--This option causes the program to display the following screen:

1. Ensure DOSIMAT, COLORIMETER and MAGNETIC STIRRER are ON.
2. Rinse COLORIMETER PROBE with DISTILLED WATER.
3. Place FLASK on STIRRER, insert MAGNET and COLORIMETER PROBE making sure PROBE SLOT is covered.
4. Place MAGNETIC STIRRER on #4 or #5.
5. Hit RETURN when ready

If five standards are already in the database, the following error message will be displayed instead:

ERROR: Maximum number of STANDARDS has been reached !

If fewer than five standards are in the database, the system will allow new standards to be titrated until a total of five is reached. It is not good practice to leave old standards in the database. These should be deleted using the 'DELETE Standards' selection of the

'(3) Edit Standards and Blanks' option on the first screen. The system uses an average of all standards in the database and therefore would be using an erroneous value if old standard values were present.

When <ENTER> is pressed, the system will start the titration, instructing the Dosimat to dispense the titrant and monitoring transmittance from the colorimeter. (Note: The program does not request a sample number or bottle number to determine standards.) The program plots transmittance % versus volume of titrant on the screen, logs both transmittance and titrant volume in a temporary file on the B: drive and carries out a titration endpoint algorithm which shuts off the Dosimat after a likely endpoint has been reached (see the beginning of Section 5). At this point the computer requests the user to confirm that the titration is complete. If for some reason the sample is still coloured and the user types 'NO', the program will set the Dosimat to Mode 2, in which a slug of titrant 1/10,000 of the burette volume, is released into the sample. The program will ask each time for a user response to indicate that the titration is complete.

When the user types 'Y' the system will signal the Dosimat to refill the burette, and will carry out an endpoint determination routine, store the endpoint in the database, and display a message with 'y/n' options asking if the user wants to do more standards. The system will commence the next titration on a 'Y' and return to the main screen menu on a 'N'. Each time a titration is completed, the system logs the volume of titrant in the database.

After triplicate standards, the user will check precision by selecting the '(3) Edit Standards and Blanks' option from the main screen.

(6) *Perform Blank Titrations*--This option differs slightly from '(5) Perform Standard Titrations' in that two titrations are carried out for each blank instead of one each as in the case of standards. Following the first titration the system sets up automatically in preparation for a second titration without input from the user. The user adds an additional amount of potassium iodate and presses <ENTER> to have the titration continue. After the second titration the system displays a message with 'y/n' options asking if the user wants to do more blanks. The system will commence the next titration on a 'Y' and return to the main screen on a 'N'.

(7) *Perform Titrations*--This option controls titration of samples. It gives instructions for setting up the titration, requests sample information, sets up headings for results on the printer, controls equipment during the titration, and then logs resulting data to diskette files and the printer.

Selection of '(7) Perform Titrations' causes the program to (1) print date and time with headings for a data table and (2) display the prompts for sample identifier information. The data table appears as follows on the printer:

Date: 1991/11/8 Time: 17:53:6:73

Sample ID	Bottle Number	Titrant Volume	Oxygen Concentration

The system then requests the following information:

What is your sample identifier?

The sample identifier must be a positive integer. Further, if you make an error and use the <backspace> to correct it, the system will respond with the following message:

Illegal input, non-numeric character. Please re-enter!

After correctly typing the number and <ENTER>, the system will display the following prompt:

What is your bottle number? (9999 for none).
---->

If a non-existent bottle number is given, the system will display the following error message and repeat the request for the bottle number:

ERROR: Bottle 245 not found!

Entering '9999' permits the system to carry out a titration and log data but omits the calculation of oxygen concentration. This option is used if a bottle does not have a number or if a bottle accidentally has the wrong stopper. Later the correct volume can be measured and entered into the system database using the '(4) Edit Bottle File' option, and the oxygen concentration can be calculated using the '(9) Recalculate Oxygens' option.

If no values are entered in response to either of these prompts, pressing <ENTER> will cause the system to display the message:

Want to do another TITRATION? (y/n)

A 'YES' response returns to the prompt requesting a new sample identifier, while 'NO' returns the user to the main screen menu.

If the user enters valid numbers for sample identifier and bottle number the system will display titration instructions as follows:

1. Ensure DOSIMAT, COLORIMETER and MAGNETIC STIRRER are ON.
2. Rinse COLORIMETER PROBE with DISTILLED WATER.
3. Place FLASK on STIRRER, insert MAGNET and COLORIMETER PROBE making sure PROBE SLOT is covered.
4. Place MAGNETIC STIRRER on #4 or #5
5. Hit RETURN when ready

This screen is the same as used in the standard and blank titrations and the steps which follow are identical to those described for '(5) Perform Standard Titrations' in the main menu. In addition the system logs the volume of titrant and computed oxygen concentrations in the database and concurrently prints the sample data under the headings printed when the option was selected.

The system displays the following prompt after it has completed the titration routine and logged resulting data:

Want to do another TITRATION? (y/n)

A 'YES' response returns to the prompt requesting a new sample identifier, while 'NO' returns the user to the first screen.

(8) *Display Endpoints*--This option displays an on-screen list of endpoints for the current round of titrations as follows:

Sample Ident	Bottle Number	Endpoint
15241	873	1.017
15245	682	1.220
15263	107	1.111

If no samples are in the current database, the message ' *** NO SAMPLES ON DISK ***' is displayed.

This option does not print out the results.

(9) *Recalculate Oxygens*--This option allows oxygen concentrations to be recalculated after changes are made to the header file or bottle file. The option leaves the data on titrant volume intact. The option also arranges the oxygen determinations by sample number in ascending numerical order. If the samples do not have to be recalculated, this is a convenient way of arranging the oxygen data.

If no samples are in the current database, the message ' *** NO SAMPLES ON DISK ***' is displayed.

(10) *Display Oxygens*--This option displays an on-screen list of oxygen concentrations in the current round of titrations as follows:

Sample Ident	Oxygen (mL/L)
15241	6.820
15245	7.210
15263	7.012

If no samples are in the current database, the message ' *** NO SAMPLES ON DISK ***' is displayed.

This option does not print out the results.

6. MAINTENANCE

Every now and then (normally about once a week) gently clean the exposed optical surfaces (lens and mirror) with a cotton swab (Q-tip and water). The probe and tube for dispensing thiosulphate should not be immersed for prolonged periods when the system isn't operating as the silver coating on the probe mirror can be damaged. The anti-diffusion tip, however, does not appear to be affected by long periods of immersion. The anti-diffusion tip has a one-way valve to prevent titrant from leaking into the sample when not under pressure from the Dosimat.

7. REFERENCES

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